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Mast Cell Stabilizing Properties of Antihistamines

Francesca Levi-Schaffer[1] and Ron Eliashar[2]

Histamine is a key mediator of allergic inflammation, primarily through competitive antagonism of binding to H1 receptors. In this issue, Weller and Maurer report that the H1 antagonist desloratadine possesses mast cell–stabilizing properties when challenged in an IgE-dependent or -independent fashion. Thus, desloratadine provides benefits that are independent of H1 receptor binding and based on mast cell stabilization.

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Histamine

Several mediators are involved in the pathophysiology of allergic diseases. Among them, histamine is key, especially in urticaria and rhinitis. Histamine was first identified as a mediator of biological functions in the early 1900s, and new roles are still being identified. Histamine interacts with the four histamine receptor subtypes: H1, H2, H3, and H4. These G-protein-coupled receptors differ in their location, second messengers, and histamine-binding properties. In allergy—from allergic rhinitis and conjunctivitis to urticaria, atopic dermatitis, and asthma—the typical histamine receptor has been thought to be H1, and the typical antihistamines have been the H1 antagonists, which have been in clinical use for more than 60 years. Recently, the role of the H4 receptor in inflammatory and allergic diseases has attracted attention (Zampeli and Tiligada, 2009).

In the skin, histamine provokes erythema, edema, and itching. In the nose, it induces itching, sneezing, edema, obstruction, and rhinorrhea. In the lungs, it is primarily a bronchoconstrictor. The symptoms of allergic inflammation (AI) are produced not only by histamine, but also by other inflammatory mediators that are released primarily by mast cells (MCs) and basophils activated by antigen–IgE interactions. Antihistamines act through the competitive antagonism/inverse receptor agonism observed in histamine's binding to H1 receptors on nerve endings, smooth muscle, and glandular cells. However, it has long been speculated that they might also possess anti-inflammatory and MC-stabilizing capabilities.

The current evidence

Weller and Maurer (2009, this issue) questioned whether desloratadine has human skin MC-stabilizing properties. Desloratadine, the main metabolite of loratadine, is a rapidly active, once-daily, nonsedating, selective high-affinity H1-receptor antagonist/inverse receptor agonist that can also interact with the five subtypes of muscarinic receptors. It has 10–20 times the *in vivo* receptor-binding affinity of loratadine and displays linear pharmacokinetics after oral administration. It is rapidly absorbed and metabolized in its first passage through the liver by cytochrome P450. Desloratadine has proven efficacy and safety in the control of AI symptoms attributable to both its anti-H1 properties and other anti-inflammatory effects.

To study the skin MC-stabilizing properties of desloratadine, the authors purified human skin MCs and then challenged them with anti-IgE antibodies or in an IgE-independent fashion with substance P or Ca-ionophore. Both histamine release and the expression of the MC-activating marker, CD107a, were evaluated after preincubation with and in the subsequent presence of desloratadine (10^{-8}–10^{-4} M). Desloratadine inhibited both MC activation and

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CD107 expression in a dose-dependent manner, with slight differences among the activators. It was therefore concluded that desloratadine could be considered an effective MC stabilizer. Previously, Genovese *et al*. (1997) demonstrated that desloratadine caused concentration-dependent inhibition of histamine, tryptase, LTC_4, and PGD_2 release from human skin MCs challenged with anti-FcεRI. However, in that study, the MCs were only partially purified, and the stabilizing effect of the drug could be attributed to an indirect effect through the contaminating cells, rather than to a direct effect on the MCs. The present study is important because it provides evidence of a direct MC-stabilizing property of desloratadine on human dermal MCs challenged with both IgE-dependent and -independent stimuli, enhancing the value of this drug in treating allergic skin diseases and non-allergic MC-associated diseases.

Anti-H1, antiallergic, and MC-stabilizing properties

Anti-H1 drugs such as azatadine, cetirizine, mizolastine, and newer -generation drugs such as desloratadine, fexofenadine, and levocetirizine have been shown to have antiallergic/anti-inflammatory and MC-stabilizing properties. The properties of these antihistamines and other drugs have been extensively summarized and reviewed by Assanasen and Naclerio (2000), Agrawal (2004), and de Benedictis *et al*. (2008). Importantly, the effects of each drug differ quantitatively and qualitatively, and they can be both receptor dependent and independent. *In vitro*, some of these drugs may reduce chemotaxis, activation, and survival of eosinophils; alter the expression of epithelial cell adhesion molecules; or alter the production of proinflammatory cytokines and the Th1/Th2 cytokine balance in basophils and T cells. *In vivo*, administering these drugs results in a variety of effects. For example, cetirizine reduces eosinophil infiltration at the site of allergen challenge in skin, but not in the nose (Michel *et al*., 1988; Klementsson *et al*., 1990). On the other hand, mizolastine inhibits antigen-induced eosinophil infiltration in mouse skin, as well as in the nasal cavity of guinea pigs, and reduces leukotriene generation (Yamada and Funayama, 1998). Mizolastine inhibits both the early and the late antigen-induced soluble ICAM in human skin blisters (Michel *et al*., 2000).

> Desloratadine benefits allergy via H1 receptor-binding and mast cell stabilization

The MC-stabilizing properties of the anti-H1 drugs are variable. Okayama and colleagues (1994) found that cetirizine inhibited IgE-dependent histamine and PGD_2 release from lung and tonsil MCs, but it exhibited weak inhibitory activity on skin MCs, which was not concentration dependent. Terfenadine, on the other hand, had a dual effect on skin MCs; at low concentrations, it exerted a concentration-dependent inhibition of IgE-dependent histamine release, whereas at higher concentrations it stimulated mediator release. Ketotifen produced similar effects (Okayama *et al*., 1994). Nielsen and co-workers (2001) observed that cetirizine inhibited *in vivo* human skin reactions but not mediator release in immediate allergic cutaneous reactions. They concluded that cetirizine is a potent H1-receptor antagonist but that it has no effect on MC activation.

Finally, as reviewed by Marshall (2000) and Agrawal (2004), desloratadine has various anti-inflammatory effects. It inhibits ICAM expression in nasal epithelial cells and P-selectin expression in umbilical vein epithelium, as well as the release of IL-8, RANTES, and soluble ICAM from human bronchial epithelial cells. Moreover, it decreases the secretion of various inflammatory cytokines from human leukemic MC and basophil lines and, in general, it downregulates many immune/inflammatory cell activities, such as those of eosinophils and B cells.

Possible mechanisms for desloratadine-induced MC stabilization

Weller and Maurer (2009) studied how desloratadine exerts its inhibitory effect on MCs. Different hypotheses exist for the mechanisms behind the MC-stabilizing properties of antihistamines; these are only suppositions, owing to a lack of experimental evidence. Notably, the mechanism of action of the "classic" MC-stabilizing drug cromolyn sodium remains ill defined. Anti-H1 drugs are lipophilic cationic compounds that can permeate cell membranes, stabilizing FcεRI on MCs and preventing the perturbations otherwise caused by immunological stimuli. However, Okayama and co-workers (1994) found no correlation between the lipophilicity of different antihistamines and their inhibitory effects on histamine release by MCs. Another mechanism of MC stabilization may be the direct interference of antihistamines with Ca^{2+} channels, intracellular Ca^{2+} release, and Ca^{2+} utilization. The dissolving of the lipophilic ends of antihistamines into cell membranes leads to the presentation of a positive charge outside of a cell, which can inhibit competitively the binding of Ca^{2+}.

Berthon and colleagues (1994) found that loratadine and desloratadine impair the increase in Ca^{2+} that follows cell activation by decreasing both the influx of extracellular Ca^{2+} and the release of Ca^{2+} from intracellular stores. The reduction of intracellular Ca^{2+} reduces the activity of Ca^{2+}-dependent enzymes, such as calmodulin. Indeed, blocking of calmodulin *in vitro* leads to inhibition of histamine release from MCs as a result of a decreased activation of enzymes essential for the secretory process. Moreover, direct inhibition of calmodulin by antihistamines may also lead to inhibition of MC-mediator release. The degree of inhibition observed with desloratadine in Weller and Maurer's study (i.e., around 60% for antigen and around 20% for Ca-ionophore) was observed in an earlier study using the calmodulin antagonist trifluoperazine (Chakravarty and Nielsen, 1985). However, to the best of our knowledge, the direct influence of antihistamines on calmodulin has not yet been studied.

Another possible mechanism of MC stabilization by desloratadine could be the inhibition of autocrine stimulation by histamine. Human skin MCs have been found to express H1, H2, and H4 but not H3 receptors (Lippert *et al*., 2004).

In theory, histamine released by MCs may bind to H1 receptors on their own surface, thereby exerting an autocrine amplification of MC-mediator release. However, in addition to the weak expression of H1 receptors on skin MCs, it has been shown that preincubation with an H1-antagonist caused an increase of histamine-induced cAMP on these cells, probably because of competitive antagonism between H1 and H2 receptors on the same cell. Moreover, histamine seems to have an inhibitory rather than an activating effect on MCs via H2 receptors in terms of histamine and cytokine release. Because histamine may exert MC-activating effects by binding to H4 receptors, antihistamines could block this receptor. Nevertheless, desloratadine—and cetirizine and fexofenadine—has shown no inhibition of the H4 receptor at concentrations up to 10 μM.

Given the above data, we are tempted to speculate that desloratadine exerts its inhibitory effects on human skin MCs by interfering with either intracellular Ca^{2+} accumulation or activation of intracellular Ca^{2+}-dependent enzymes.

The concentrations of desloratadine required to obtain significant MC stabilization in this *in vitro* study (10^{-5}–10^{-4} M) are higher than those found in blood (around 10^{-8} M). It is possible that MC-stabilizing effects *in vivo* will be achieved only at higher doses. Nevertheless, Frossard and colleagues (2008) were recently able to detect more than 100-fold higher concentrations of desloratadine in skin than in plasma.

Conclusions

It is clear that desloratadine and other H1 antagonists provide beneficial effects in AI that are not directly H1 receptor binding–linked and are generally based on anti-inflammatory properties and inhibition of MC/basophil histamine release.

Weller and Maurer's study supports an *in vitro* (and possibly *in vivo*) role for desloratadine as a skin MC stabilizer. The mechanisms by which desloratadine and other antihistamines act as MC stabilizers are not yet identified, but they are likely concentration dependent and diverse. Ad hoc *in vitro* mechanistic studies are needed to define the MC-stabilizing properties of these drugs.

The *in vitro* concentrations of desloratadine required to achieve MC stabilization are considerably higher than those achieved at therapeutic doses. It has not been established whether the antiallergic/anti-inflammatory properties described *in vitro* and in animal models exist in humans, nor has the mechanism of action or its clinical significance been determined. To provide a definite answer, the antiallergic/anti-inflammatory and MC-stabilizing properties of antihistamines should be demonstrated *in vivo*, in allergic patients, and at therapeutic and safe doses.

CONFLICT OF INTEREST

The authors state no conflict of interest.

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